

LS-174T growing culture | 330392

General information

Description	The line was derived from the same tumor as LS 180. LS 174T cells stain positively for cytokeratins.
Organism	Human
Tissue	Colon
Disease	Adenocarcinoma
Synonyms	Ls174T, LS174t, Ls-174-T, LS-174-T, LS 174 T, LS174T, Ls-174T, LS 174T, LS-174, LS174

Characteristics

Age	58 years
Gender	Female
Ethnicity	Caucasian
Morphology	Epithelial-like
Growth properties	Adherent

Identifiers / Biosafety / Citation

Citation	LS-174T (Cytion catalog number 300392)
Biosafety level	1

Expression / Mutation

Protein expression	Colon Antigen 3 +, CEA +, p53 -, GFAP -, mRNA expression +
Antigen expression	HLA A2, B13, B50, Blood type O
Isoenzymes	ADA, 1: G6PD, B, PGM1, 1, PGM3, 2, PGD, A, ES-D, 1, PEP-D, 1

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Oncogenes	myc +, myb +, ras +, fos +, p53 +, sis -, abl -, ros -, src -
Tumorigenic	Yes, in nude mice
Reverse transcriptase	negative
Products	Carcinoembryonic antigen (CEA) 1944 ng/106 cells in 10 days, mucin, interleukin-10 (IL-10), interleukin-6 (IL-6)
Mutational profile	LS-174T cells carry a mutation in codon 12 of Kras gene: GGT(Wt Gly) >GAT(Asp)
Karyotype	45,x with one x chromosome missing but no other chromosomal aberrations
Handling	
Culture Medium	EMEM, w: 2 mM L-Glutamine, w: 1.5 g/L NaHCO ₃ , w: EBSS, w: 1 mM Sodium pyruvate, w: NEAA (Cytion article number 820100c)
Medium supplements	Supplement the medium with 10% FBS
Passaging solution	Accutase
Subculturing	Remove medium and rinse the adherent cells using PBS without calcium and magnesium (3-5 ml PBS for T25, 5-10ml for T75 cell culture flasks). Add Accutase (1-2ml per T25, 2.5ml per T75 cell culture flask), the cell sheet must be covered completely. Incubate at ambiente temperature for 8-10 minutes. Carefully resuspend the cells with medium (10 ml), centrifuge for 5 min at 300xg, resuspend cells in fresh medium and dispense into new flasks which contain fresh medium.
Split ratio	A ratio of 1:2 to 1:5 is recommended
Seeding density	5 to 8 x 10 ⁴ cells/cm ²
Fluid renewal	2 to 3 times per week
Freezing recovery	After thawing, plate the cells at 5 x 10 ⁴ cells/cm ² and allow the cells to recover from the freezing process and to adhere for at least 24 hours.
Freeze medium	CM-1 (Cytion catalog number 800100) or CM-ACF (Cytion catalog number 806100)

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Handling of cryopreserved cultures

LS-174T cells are shipped in a deep-frozen state on dry ice. Upon receipt, confirm that the vial remains frozen. For storage, place the cryovial immediately at temperatures below -150 degrees. If you plan to culture the cells immediately, swiftly thaw the vial by shaking it in a 37 degrees water bath with clean water and an antimicrobial agent for 40-60 seconds. Remove the vial once a small ice clump persists, ensuring it remains cold. Proceed with all subsequent steps under aseptic conditions. In a sterile flow hood, disinfect the cryovial with 70% ethanol. Then, gently open the vial and transfer the cell suspension into a 15 ml centrifuge tube pre-filled with 8 ml of room temperature culture medium. Gently mix the cells. For cell separation, centrifuge at 300 x g for 3 minutes and dispose of the supernatant. Skipping centrifugation is optional, although any residual freezing medium should be removed after 24 hours. Resuspend the pellet gently in 10 ml of fresh culture medium and divide between two T25 culture flasks. Follow the subculture protocol for subsequent steps.

Handling of proliferating cultures

One or two cell culture flasks come filled with cell culture medium. Collect the entire medium in a 50 ml centrifuge tube. Spin down the collected medium at 300 x g for 3 minutes to collect the cells which may have detached during transit. If a cell pellet is visible, resuspend the cells in 5 ml of cell culture medium and transfer to a T25 cell culture flask. Carefully add 5 ml of cell culture medium to each T25 cell culture flask. Examine cell morphology and confluency using a microscope. Finally, incubate the flasks at 37 degrees Celsius for at least 24 hours.

Quality control / Genetic profile / HLA

Sterility

Mycoplasma contamination is rigorously excluded using both PCR-based assays and luminescence-based mycoplasma detection methods. To ensure there is no bacterial, fungal, or yeast contamination, cell cultures are subjected to daily visual inspections.

STR profile

Amelogenin: x,Y
CSF1PO: 10,14
D13S317: 10,11
D16S539: 11,13
D5S818: 11,15
D7S820: 10.3,11
TH01: 6,7
TPOX: 8,9
vWA: 15,17,18,19
D3S1358: 15,17
D21S11: 29,30,31
D18S51: 11,13
Penta E: 15,16
Penta D: 10
D8S1179: 11,12,16
FGA: 21,22
D1S1656: 12,13,14,18.3,19.3
D6S1043: 12,13,14
D2S1338: 18,22
D12S391: 18,19,20
D19S433: 13,14,15

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HLA alleles

A*: 02:xx, 30:01:01

B*: 13:xx, 35:01:01

C*: 04:01:01, 06:xx

DRB1*: 04:02:01, 07:01:01

DQA1*: 02:01:01, 03:01:01

DQB1*: 02:02:01, 03:02:01

DPB1*: 03:01:01G, 04:01:01

E: 01:01, 01:03